filled bubbles within a sponge like gelatin matrix. In vitro ultrasonic testing revealed a reflected digital echo intensity comparable to that of air filled albumin microbubbles. However, the *microsponge* digital ultrasound intensity persisted considerably longer during ultrasound exposure; *microsponge* intensity half-life 200 sec, albumin microbubble half-life 2 sec. In vivo testing in 4 dogs revealed left heart and myocardial *microsponge* persistence half-times of 200 to 300 sec compared to albumin microbubble half-times of 3–5 sec. Thus a newly developed *microsponge* echo-contrast agent produces a bright contrast echo effect and appears to resist pressure and ultrasonic collapse. This new agent shows a 100 fold greater myocardial and left heart persistence. These *microsponges* considerably enhance myocardial perfusion studies.

## 1004-109 Antigen Binding Characteristics of Antibodies Coupled to Targeted Acoustically Reflective Immunoliposomes

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We have demonstrated acoustically reflective liposomes targeted successfully to fibrin-bearing structures by covalent coupling to polyclonal rabbit IgG antibodies that react equally with fibrinogen and fibrin (Am Diagnostica, #313R). Enhanced acoustic imaging was demonstrated in vitro and in vivo using a miniswine model of induced atherosclerosis. To determine the effective antibody affinity required for successful liposome targeting, the ability of the antibody preparation to increasing bind $I^{125}$ fibrinogen in the presence of increasing quantities of unlabeled fibrinogen (double-antibody radioimmunoassay protocol) was assessed at three temperatures (4, 24 and 37ºC).

*Results:* Two major orders of binding affinity were found: high ($10^8$ $M^{-1}$) and low ($6 \times 10^6$–$6 \times 10^7$ $M^{-1}$). Higher affinity antibodies showed moderate negative enthalpy and high positive entropy, indicating hydrophobic interactions whereas lower affinity antibodies showed high negative enthalpy and moderate negative entropy, indicating ionic interactions.

*Conclusions:* Our results suggest that acoustically reflective liposomes can be targeted for site specific imaging using low-affinity antibodies with major implications for their use for multistructure targeting, acoustic enhancement and drug delivery.

## 1004-122 Safety and Efficacy of AF0150 for Cardiac Contrast Enhancement in Normal Subjects: Initial Results of a Phase I Clinical Trial

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AF0150 (Imagent® US, Alliance Pharmaceutical Corp.) is a new perfluorohexane-stabilized ultrasound contrast agent that have demonstrated to produce intense LV cavity and myocardial opacification following IV injections in animals. To determine the clinical safety and efficacy of this agent, a Phase I study was undertaken in 64 healthy volunteers. 20 subjects received placebo and 44 received incremental doses of AF0150 (0.125, 0.50, 2.0 and 4.0 mg/kg. Cardiac imaging was obtained with commercial instruments at 3.5 MHz in the parasternal short axis or apical views. Safety assessments included BP, HR, ECG, ventilatory status, $O_2$Sat., CBC, chemistries, coagulation and urine analysis. All subjects tolerated all the injections without adverse events and no clinically significant changes in the parameters obtained were observed. Echo revealed, at every dose, dense and total opacification of the LV cavity following transient cavity and myocardial shadowing. Myocardial opacification was visible in non-shadowed segments after all injections in each subject. Videodensitometry, 4 mm radius ROI positioned in the mid wall of non-shadowed segments revealed an increase from baseline of 21 to 68, 40 and 49 gray levels at the incremental doses of 0.125, 0.50 and 2.0 mg/kg, respectively. Thus AF0150 produces dense LV cavity and myocardial opacification in normal subjects without significant adverse effects or clinical or laboratory abnormalities. This agent should prove useful for the clinical assessment of myocardial perfusion.

## 1004-123 Influence of Ultrasonic Energy on Contrast Echocardiography: Intermittent Imaging Using AF0150 Yields Generalized Myocardial Opacification While Continuous Imaging Delineates Intramyocardial Vessels

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AF0150 (Imagent®, Alliance) is an intravenous contrast agent that has been found to produce linear or punctate signals compatible with intramyocardial vessels rather than a diffuse myocardial blush during second harmonic (2H) imaging. We have demonstrated that bubble destruction by acoustic energy was increased during low velocity flow. We hypothesized that continuous (CONT) insonification destroyed bubbles in the low velocity microcirculation but not in higher velocity vessels, and that this phenomenon could be reversed by intermittent imaging. Therefore, we compared CONT with ECG-gated imaging in 8 dogs given 1.5 mL of AF0150 IV. Short axis echo at the mid-papillary level was performed using different instruments capable of 2nd H imaging by transmitting from 1.8 to 2.5 MHz and receiving between 3.6 and 5.0 MHz. Pulsed Doppler recordings were obtained from the linear structures. Off line videodensitometry (4 $mm^2$ ROI, 0–255 gray levels) was done on the linear structures and adjacent myocardium. CONT imaging produced linear or punctate structures compatible with intramyocardial vessels with a mean intensity of 96.8 ± 55 gray levels (mean ± SD), while myocardial intensity was the same as baseline: 30.8 ± 17 GL. Pulsed Doppler recordings from these linear structures yielded predominantly diastolic velocities compatible with coronary flow. ECG-gated imaging yielded a diffuse myocardial blush with a mean peak videointensity of 93.4 ± 35 GL throughout the wall. Thus, during 2H imaging of AF0150, continuous insonification visualizes intramyocardial vessels while intermittent imaging yields a generalized myocardial blush. These data support the concept that lower velocity bubbles in the microcirculation are destroyed by acoustic energy to a greater extent than those in vessels. This differential response may be of clinical value.

## 1004-124 Studies of Bubble Persistence vs Standard and Harmonic Mode Acoustic Pulse Pressure for Three New Echocontrast Agents

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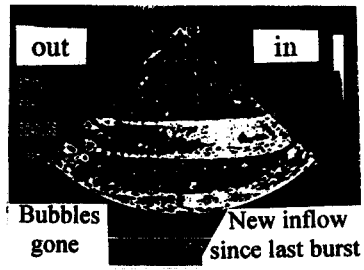
We developed a non-recirculating, pressurized in vitro flow model in which infused contrast echo bubbles flow through a gelatin matrix at a constant flow rate. Ultrasound imaging was performed with an HP SONOS 2500 prototype allowing us to control and to equalize pulses per burst and power output for standard and harmonic imaging. For continuous or intermittent gated exposure, measurements by on-line Acoustic Densitometry and bubble counts using an optical Acusizer™ m770A system were obtained at inlet and outlet. Three agents were studied: MRX115 (Aerosomes, ImaR); SonoGen (QW7437, Sonus Pharm.); and Imagent (AFO 150) (Alliance Pharm.). MRX showed a relative threshold for bubble destruction which increased above a transmit gain of 40 (5.1 watts/$cm^2$ spatial peak pulse average). At levels above this, even gated single burst imaging could destroy all bubbles already present within the imaging field (Fig.).




The Alliance agent showed similar characteristics with a destruction threshold between 1 and 5 watts/$cm^2$. There was no difference between standard and harmonic imaging results for either agent once pulse power output and pulses per burst were equalized. The Sonus agent, which continued to generate bubbles in our model at 37º, showed more variability, but once stabilized, its sensitivity to ultrasound power paralleled results for the other agents. Our unique model combines controlled ultrasound power and exposure as well as optical particle counting to provide insights about the interaction between ultrasound imaging and bubble persistence.